Table 8.1. The CNS and QM/MM x-ray refinement of the C1, C2, and C3 conformers of 1,6-dihyroxynaphthalene bound to Orf2

Conformers	Refinement protocol	X-ray weights	R	R_{free}	Distance (Å)	
					D1	D2
C1	QM/MM	0.01	0.2540	0.2674	3.96	7.09
		0.2	0.2419	0.2629	3.97	7.12
		1.0	0.2290	0.2628	4.01	7.21
	CNS	0.01	0.3735	0.4015	5.03	8.17
		0.2	0.2606	0.3004	4.53	7.71
		1.0	0.2307	0.2754	4.10	7.17
C2	QM/MM	0.01	0.2604	0.2894	6.89	9.82
		0.2	0.2432	0.2798	6.78	9.73
		1.0	0.2285	0.2734	6.80	9.79
	CNS	0.01	0.3690	0.4021	8.15	10.48
		0.2	0.2617	0.3015	7.53	10.28
		1.0	0.2320	0.2763	7.10	10.11
C3	QM/MM	0.01	0.2496	0.2795	5.91	4.04
		0.2	0.2414	0.2749	5.81	3.96
		1.0	0.2283	0.2699	5.87	3.97
	CNS	0.01	0.3709	0.4018	7.36	5.27
		0.2	0.2642	0.3057	6.95	4.56
		1.0	0.2315	0.2777	6.42	4.20

CNS refinement (using a classical E_{chem} term) is shown in Table 8.1 and Figure 8.3. In Table 8.1 the weights and the resulting value of R and R_{free} (indicators of the refinement quality where lower is better) for the refinement of the three structures using QM/MM and CNS indicate that the latter refinement is superior to the former for all weights used. The results from this study show the possible improvements in structure quality possible with a QM/MM refinement, but further validation is required on other protein/ligand systems.

The development of high-throughput crystallography has called for improvement of the conventional refinement methods. Recently, Schiffer et al. reviewed the latest advances in simulation techniques that would affect the field of protein crystallography, and the use of QM methods was recognized as one of the three major forefronts.[26] With the capability and efficiency of QM continuing to increase, development and application of QM-based x-ray refinement methodologies will present many new interesting possibilities.

NMR refinement of protein/ligand complexes

Over the past decade, NMR spectroscopy has proven to be a powerful and versatile tool for the study of protein/ligand interactions. The three-dimensional structures of protein/ligand complexes can be determined by combining interproton distance restraints derived from the nuclear Overhauser effect (NOE) with other restraints from J coupling constants, hydrogen bonds, and/or residual dipolar couplings. Up to November 2006, there were over 800 NMR structures of protein/ligand complexes deposited in the Protein Data Bank. However, this determination process is far from automated and high-throughput because it is difficult to obtain accurate NMR restraints. Because Fesik and coworkers introduced SAR (structure/activity relationship) by NMR,[27] many NMR-based screening methods have been developed to identify potential drug molecules in pharmaceutical research (for reviews, see Homans, Lepre et al., and Meyer and Peters).[28–30] A recent interesting application of NMR-based screening methods is to predict protein druggability.[31,32] All these techniques take advantage of the fact that on ligand binding, significant perturbations can be observed in NMR parameters of either the receptor or the ligand. These perturbations can be used qualitatively to detect the complex formation or quantitatively to measure the binding affinity.

Among these NMR parameters, chemical shifts are exquisitely sensitive on the chemical environments of compounds. Therefore, theoretical calculations of chemical shift perturbations (CSP) on ligand binding can provide more insights about protein/ligand interactions at